The PsbW protein stabilizes the supramolecular organization of photosystem II in higher plants

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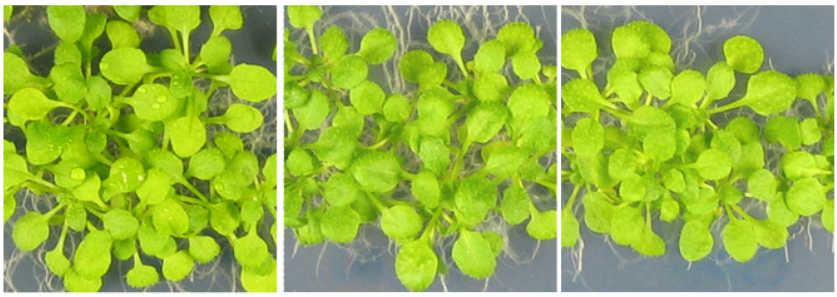
Supporting Information

Supplementary Figure S1



WT koPsbW asPsbW

Figure S1

Photograph of wild-type Col-O (WT), PsbW knock-out (koPsbW) and PsbW antisense (asPsbW) plants grown in Murashige and Skoog media agar plates showing that deleting or decreasing PsbW has no visible effect on phenotype.

Supplemental figure 2

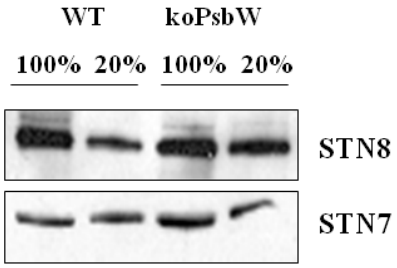


Figure S2

Immunodetection of protein kinases STN7 and STN8 in thylakoid membranes of dark-adapted leaves harvested from wild-type and koPsbW. 100% corresponds to 5 µg of chlorophyll.

Supplementary Figure S3

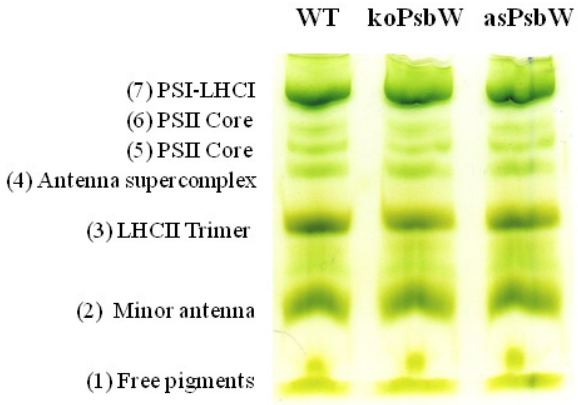


Figure S3

Characterization of the thylakoid membrane chlorophyll-containing protein complexes isolated from WT, asPsbW and koPsbW. Separation of the protein complexes in the thylakoid membrane by non-denaturing Deriphat-PAGE. Bands were identified according to (de Bianchi et al., 2008). Loading corresponds to 15 µg of total chlorophyll.

Table S1

The photosynthetic properties of koPsbW plants were examined in more detail by measuring other fluorescence parameters (Maxwell and Johnson, 2000), including the operational PSII quantum yield (ΦPSII), photochemical quenching (qP) and non-photochemical quenching (NPQ) (Table 1). ΦPSII defines the efficiency to use absorbed light energy for PSII photochemistry under continuous illumination. Under photon flux densities of 100 and 500 μmol m^{-2} s^{-1}, ΦPSII was slightly lower in the koPsbW plants than in wild-type (Table 1), possibly due to a slower rate of PSII linear electron transport in the mutant. However, the qP parameter, indicating the proportion of open PSII reaction centers under continuous illumination, was not considerably different in the koPsbW plants compared to wild-type. Similarly, NPQ, which is related to the antenna heat dissipation, was unchanged in koPsbW compared with wild type plants under these conditions as well as under illumination of 900 μmol m^{-2} s^{-1} (data not shown).

	Wild-type		**koPsbW**	
Fluorescence Illumination (PAR)[1]	100	500	100	500
ΦPSII	0.73 ± 0.01[2]	0.52 ± 0.01	0.69 ± 0.01	0.47 ± 0.05
qP	0.95 ± 0.01	0.78 ± 0.03	0.95 ± 0.01	0.78 ± 0.02
NPQ	0.26 ± 0.02	0.91 ± 0.07	0.23 ± 0.01	0.96 ± 0.30

[1]Illumination units are μmol photons m^{-2} s^{-1}.

[2]Errors reported are plus and minus 1 standard deviation (n=5).

Table S2

EPR analysis of the electron transport components of PSII showed that neither Tyr_D^{ox} nor the amount of oxidized and reduced Cytb559 is changed in koPsbW plants. However, the $Q_A^-Fe^{2+}$ interaction signal was decreased, in agreement with the thermoluminescence analysis (Figure 6). Interestingly, the S_2 state multiline EPR signal of the water-oxidizing complex showed significant (50%) differences in amplitude between wild-type and koPsbW preparations, most probably due to the double effect of less reducible Q_A and more mixed S state composition in the mutant before the signal induction.

	Wild-type	**koPsbW**
EPR measurements[1]		
Tyr_D^{ox}	100%	92%
S_2 state multiline signal	100%	47%
$Q_A^-Fe^{2+}$ signal	100%	72%
$Cytb_{559}$ ox/red	40/60%	45/55%

[1]Induction and measurements of EPR signals were performed according to (Mamedov *et al.*, 2008). $TyrD_{ox}$ signal is normalised against chlorophyll, whilst the S2 state multiline and $Q_A^-Fe^{2+}$ signals were based on Photosystem II ($TyrD_{ox}$). Oxidized (ox) and reduced (red) represent the low and high potential forms of $Cytb_{559}$ respectively.

Experimental error in the measurements is ± 5%.